Product Specification

5’-Amino C12 labeled Oligo d(T)

Oligo d(T) Amino C12 Labeled Primers

For research use only. Not for use in diagnostic procedures for clinical purposes. Commercial licenses may be obtained directly from Gene Link.

Shipping Condition: Ambient Storage: -20°C

Quantity	**25 µg**
Shipping Condition	**Lyophilized at ambient**
Storage	**-20°C**

Item	Catalog No	Product Name	MW	nmols
❑	26-4100-02	5'- Amino C12-Oligo d(T)12-18	4764.3*	5.2
❑	26-4112-02	5'- Amino C12-Oligo d(T)12	3851.7	7
❑	26-4113-02	5'- Amino C12-Oligo d(T)13	4155.9	6.5
❑	26-4114-02	5'- Amino C12-Oligo d(T)14	4460.1	6
❑	26-4115-02	5'- Amino C12-Oligo d(T)15	4764.3	5.5
❑	26-4116-02	5'- Amino C12-Oligo d(T)16	5068.5	5.2
❑	26-4117-02	5'- Amino C12-Oligo d(T)17	5372.7	5
❑	26-4118-02	5'- Amino C12-Oligo d(T)18	5676.9	4.7
❑	26-4119-02	5'- Amino C12-Oligo d(T)19	5981.1	4.4
❑	26-4120-02	5'- Amino C12-Oligo d(T)20	6285.3	4.1
❑	26-4121-02	5'- Amino C12-Oligo d(T)21	6589.5	3.8
*An average molecular weight is reported				

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Description

The Oligo d(T) primers of varying sizes are individually synthesized with an C-12 amino group at the 5' end that is used to conjugate to succinimidyl esters dyes, affinity ligands like digoxigenin or enzymes. Oligo dT is used primarily to prime synthesis by reverse transcriptase of the first strand cDNA using mRNA as a template for use in microarray hybridization protocol.

Oligo purity is greater than 98% as determined by denaturing polyacrylamide gel electrophoresis.

Succinimidyl ester Conjugation

Follow protocol supplied by NHS ester compound manufacturer.

Reconstitution

Recommended reconstitution is at a concentration of 50 µM (50 pmol/ µl) in RNase-free DEPC treated water or 10mM Tris pH 8.0. The stock solution can be further diluted to an appropriate working concentration as required.

To prepare a 50 µM solution of primer, use the nmol value of the lyophilized oligo and multiply by 20 to determine the volume of diluent in microliters to add.

Formula:
"Total nmol" x 20 = µl of diluent to add.

- Spin the tube briefly to bring down the contents of the tube that may have lodged in the cap during shipment. Pellet may be very small and not visible.
- Add appropriate amount of RNAse free water or 10mM Tris pH 8.0 directly to the tube. Vortex briefly.
- The above solution is 50µM. This is equivalent to 50 pmol/µl.

Fluorescent-labeled probes should be protected from light to avoid photo bleaching.
Store at -20^{o}C or below after reconstitution.

Recommended Usage

Use 2 µl of the 50 µM solution for 1 µg poly $(A)^{+}$ RNA as a template in a 20 µl reaction volume. See reaction conditions for more details.

Quality Control Data

This product is certified to prime first strand cDNA synthesis reaction using poly $(A)^{+}$ RNA as a template.

Functional Assay Conditions

The conditions given below have been tested to yield first strand cDNA synthesis and is given as an example. Variations and other protocols have been used by other laboratories using this product to yield excellent first strand synthesis. Investigators can substitute their own reaction conditions.

The quality of RNA is very important for the reverse transcription reaction. It is essential to have intact full length RNA as the template material that is free of even trace amounts of RNases and contaminating chemicals. Poor quality RNA template is usually the cause of truncated and incomplete cDNA products.

Add components in the order given below. Reaction volume can be scaled up.

Component	Volume	Comments
poly(A)$^+$ RNA in sterile water Quantity ~1.0 µg	up to 10 µl	Use RNAse free reagents and disposables.
RNase-free water	variable	Calculate total volume and add appropriate volume of RNase-free water at this stage.
50 µM oligo(dT)12-18 primer solution (50 pmol/µl = ~0.5 µg/µl)	2 µl	Final concentration is 5 µM (5 pmol/ µl).
Heat mixture to 70°C for 10 min, and quick chill on ice.		
5X first strand buffer [250 mM Tris-HCl (pH 8.3), 375 mM KCl, 15 mM MgCl2]	4 µl	
0.1 M DTT	2 µl	
dNTPs (5 mM each dNTP)	2 µl	Final concentration is 0.5 mM of each dNTP.
[α-^{32}P]dCTP (1 µCi/µl)	1 µl	Tracer optional. Add only if required.
Reverse transcriptase; 200 units	1- 2 µl	
Total Volume	**20 µl**	

Incubate at 37°C for 1 hour.

Related Products

Gene Link stocks various oligo dT primers, oligo dT VN primer, Oligo dT T7 primer, random primers, including an array of fluorescent dye labeled primers for genetic analysis using florescent detecting instruments. The C-12 amino labeled primers are ready to be conjugated to the investigators choice of NHS-activated ligand.

Random Primers are a mixture of oligonucleotides representing all possible sequence for that size. Random Primers can be used to prime synthesis in oligo-labeling similar to using hexamers and cDNA synthesis. Random prime labeling yields high specific activity labeled DNA probe which can be used for all southern, northern and in situ hybridization studies. Random Primers can be also used similar to using hexamers in cDNA synthesis in combination with oligo dT to yield more 5' end cDNA sequence.